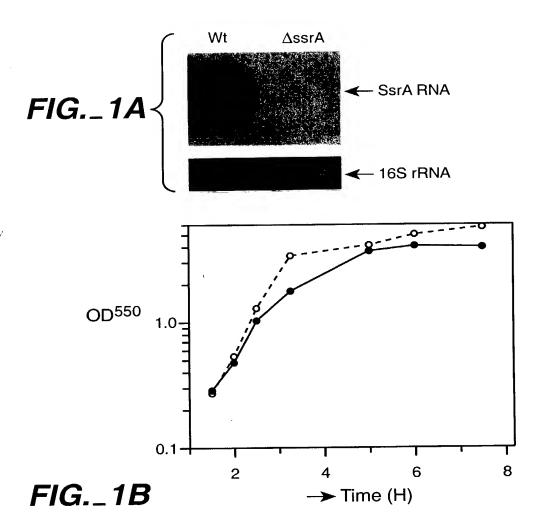
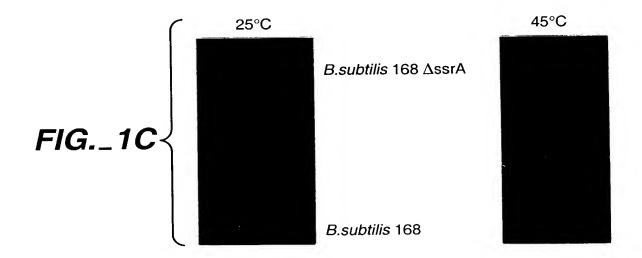
Enhanced Secretion of a Polypeptide by a
Microorganism
Marc Kolkman
SN# 09/975,132
Docket No. GC636-2
Sheet 1 of 5







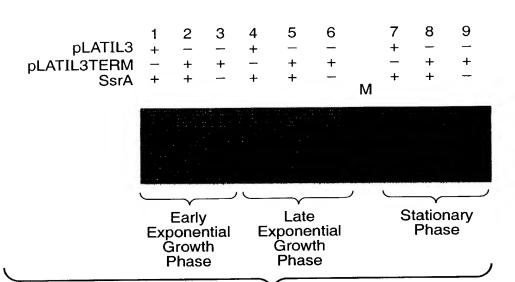


FIG._2



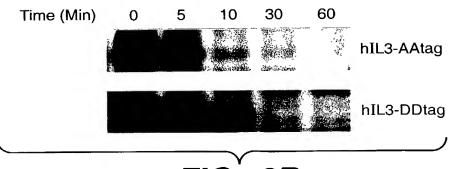
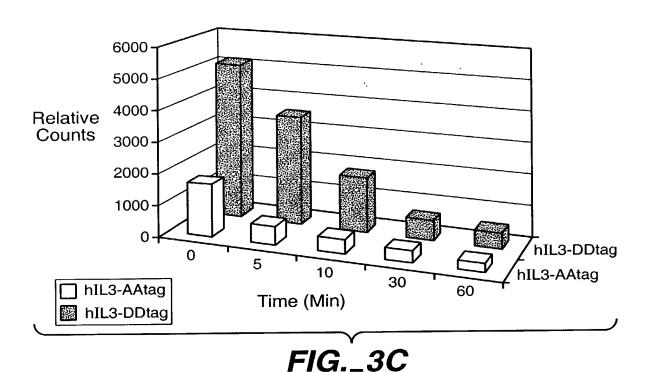


FIG._3B

Enhanced Secretion of a Polypeptide by a
Microorganism
Marc Kolkman
SN# 09/975,132
Docket No. GC636-2
Sheet 3 of 5



3 7 8 9 10 2 5 6 1 168 WB600 WB600 WB600 168 WB600 WB600 WB600 168 168 Strain AA DD DD AA AAAΑ AA SsrA AA pLATIL3 + + pLATIL3TERM SsrA-tagged hIL-3→ Run-off hIL-3→ hIL-3→ Anti-Bs-SsrAtag Anti-hIL-3

FIG._4



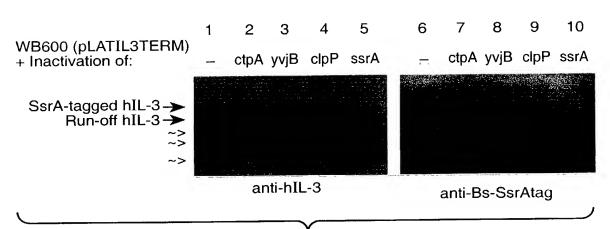


FIG._5

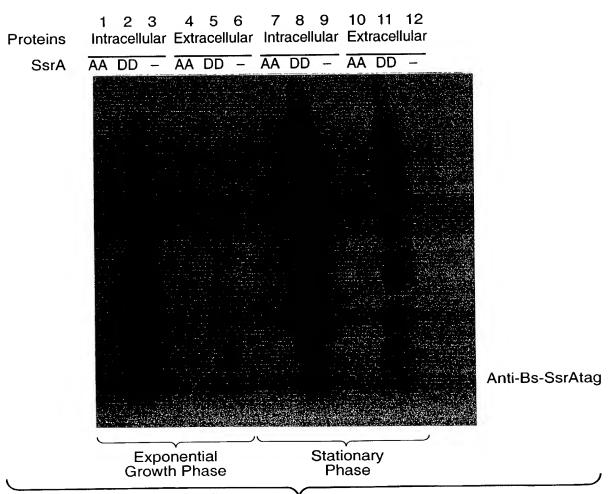


FIG._6

Enhanced Secretion of a Polypeptide by a Microorganism
Marc Kolkman
SN# 09/975,132
Docket No. GC636-2
Sheet 5 of 5



5/5

IL-3 amino acid sequence (Native sequence associated with its native signal sequence)

MSRLPVLLLL	QLLVRPGLQ A	PMTQTTPLKT	SWVNCSNMID
EIITHLKQPP			
LPLLDFNNLN	GEDQDILMEN	NLRRPNLEAF	NRAVKSLQNA
SAIESILKNL			
LPCLPLATAA	PTRHPIHIKD	GDWNEFRRKL	TFYLKTLENA
OAOOTTLSLA	IF		

IL-3 as encoded by plasmid pLATIL3

MKQQKRLYAR LLTLLFALIF LLPHSSASAA PMTQTTPLKT SWVNCSNMID EIITHLKQPP LPLLDFNNLN GEDQDILMEN NLRRPNLEAF NRAVKSLQNA SAIESILKNL LPCLPLATAA PTRHPIHIKD GDWNEFRRKL TFYLKTLENA QAQQTTLS

IL-3 amino acid sequence (Substituted tag charged C-terminus)

MKQQKRLYAR LLTLLFALIF LLPHSSASAA PMTQTTPLKT SWVNCSNMID EIITHLKQPP LPLLDFNNLN GEDQDILMEN NLRRPNLEAF NRAVKSLQNA SAIESILKNL LPCLPLATAA PTRHPIHIKD GDWNEFRRKL TFYLKTLENA QAQQTTDD

IL-3 amino acid sequence (Tagged¹⁴)

MKQQKRLYAR LLTLLFALIF LLPHSSASAA PMTQTTPLKT SWVNCSNMID EIITHLKQPP LPLLDFNNLN GEDQDILMEN NLRRPNLEAF NRAVKSLQNA SAIESILKNL LPCLPLATAA PTRHPIHIKD GDWNEFRRKL TFYLKTLENA QAQQTTLSAG KTNSFNQNVA LDD

FIG._7